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## Coagulase-negative staphylococci in blood cultures

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### Summary

Nosocomial infections with coagulase-negative staphylococci (CNS) are an increasing problem in hospitalized patients, especially in those who are immunocompromised. About 15% of the bacterial strains isolated from blood cultures are CNS. It is, however, difficult to determine if a CNS isolated from a blood culture has to be considered as a contaminant or as pertaining to bacteraemia, a difficulty not yet overcome.

The genus *Staphylococcus* is very heterogeneous and currently consists of over 30 species. In some situations, for instance in cases in which bacteraemia plays a role, it is desirable to know the name of the species. This determination can be carried out by means of commercial tests. However, these tests are expensive and lack specificity. About 75% of the strains of CNS isolated from blood cultures is *Staphylococcus epidermidis*. With a few simple and inexpensive tests, desferrioxamine susceptibility and sugar fermentation, this species can be discriminated from the other CNS (chapter II).

Some strains of CNS, especially *Staphylococcus epidermidis* again, have the property to produce extra-cellular slime. Together with other cell components this substance is able to form a biofilm, for instance on prosthetic devices, in which bacteria can protect themselves against host defence mechanisms and the action of antimicrobial agents. We have investigated the possibility to use the slime producing property for discriminating contaminants from strains isolated from blood cultures of patients with bacteraemia. There was a significant difference in the number of slime producing strains between "contaminants" and strains pertaining to "bacteraemia". However, within the group of "contaminants" a considerable number of CNS was positive, making the test less suitable for the above mentioned aim. The slime producing strains in our investigation were significantly more resistant to cloxacillin, aminoglycosides, trimethoprim, erythromycin and ciprofloxacin. It is very likely that the slime producing ability of these strains provides them with an opportunity to escape from the action of antibiotics, thus increasing the possibility to develop resistance under the continuous pressure of antimicrobial agents (chapter III).

As mentioned before, the genus *Staphylococcus* accounts for more than 30 species. Furthermore, within a species many types can be distinguished by means of fingerprinting with pulse-field gel electrophoresis (PFGE). From the 162 strains of CNS recovered from blood cultures pertaining to "bacteraemia" in a period of three years (1990-1992) 19 strains, all *Staphylococcus epidermidis*, were strongly slime positive with an optical density of  $>1.00$  in the quantitative microtiterplate test with crystal violet (chapter III). Fingerprinting with PFGE revealed that 14 strains had the same profile. Three strains had two and one strain had three bands difference and these strains can be considered as subtypes. Thus, 18 out of 19 strains recovered from blood

cultures submitted to our laboratory by different departments in a period of three years, in all likelihood progenated from the same parent strain. Because of its properties concerning slime production and resistance to antimicrobial agents the strain was able to give rise to clonal spread throughout the entire hospital (chapter IV).

Because most of the CNS are resistant to penicillin, the drug of first choice in cases where bacteraemia with CNS has to be treated, used to be cloxacillin, a small spectrum anti-staphylococcal agent. However, strains of CNS, especially *Staphylococcus epidermidis*, have become more resistant. A cloxacillin resistant staphylococcus has to be considered resistant to all penicillins, cephalosporins and carbapenems. This causes the physician to prescribe an agent like vancomycin, an expensive drug with considerable side-effects. Thus it is important to know, in a fast and reliable way, the susceptibility of a strain to cloxacillin. The best technique to determine this susceptibility is the polymerase chain reaction (PCR). However, many of the routine laboratories do not have the opportunity to explore this expensive technique and have to rely on more conventional methods. The conditions for these methods have been under investigation for many years. The conditions we used in this investigation, Mueller Hinton Agar with 2% NaCl, a 5 µg oxacillin disk and 24 hours of incubation at 35°C, yielded a good sensitivity and specificity compared with PCR (chapter V).

Strains of CNS become more and more resistant in the hospital under the continuous pressure of the use of antibiotics for therapy and prophylaxis. In chapter VI the increasing resistance to cloxacillin and aminoglycosides is demonstrated. In the department of haematology, where ciprofloxacin was used as a prophylactic agent, there was a rapid development of resistance to this agent. This was followed by a small recovery, when the prophylactic use of ciprofloxacin was diminished.

The results of this investigation are based on laboratory findings. The outcomes of this study can and should be used for further investigation on CNS in clinical trials in order to find parameters to discriminate contaminants from strains associated with bacteraemia, to define epidemiological relevance and to give directives for antibiotic policy.

Samenv

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